between decreased antioxidants and increased levels of cell alterations due to oxidative damage, supporting the idea that there is a persistence of oxidative stress in these haematological malignancies. The difference noted between the two groups could be associated with the protocol for treatment. For this reason we intend to continue the study in children just diagnosed compared to different phases of treatment, remission induction and remission maintenance.

YSF–80
The role of calcium ions in lipid bodies life-cycle of marine bacterium Alcanivorax borkumensis

Z. Purkrtova[1], B. Kralova[1] and J. Sabirova[2]
[1]*Department of Biochemistry and Microbiology, Institute of Chemical Technology, Prague, CZECH REPUBLIC,* [2]*Faculty of Bioscience Engineering, Laboratory of Industrial Microbiology and Biocatalysis, Ghent University, Ghent, BELGIUM*

Alcanivorax borkumensis is an ubiquitous marine *hydrocarbonoclastic bacterium* with ability to degrade an exceptionally broad range of alkane hydrocarbons but only few other substrates. Its lipid metabolism is characterised by production of three lipid groups (triacylglycerols, polyhydroxyalkonates and free fatty acids) that are deposited inside the cell in the form of lipid bodies. Its metabolism makes *A. borkumensis* an interesting tool for several biotechnological applications including production of biodegradable plastics and bioremediation of petroleum oil contamination in marine ecosystems. Ca^{2+} play an important role in the life-cycle of lipid bodies in several organisms (e.g. mammalian and plants), mainly during biogenesis and mobilisation phase. For that reason the evidence of calcium presence on the surface of lipid bodies of *A. borkumensis* evoked the question of potential calcium role in their life-cycle. We use several approaches, such as protein profile determination and lipid analysis, to elucidate this role. Our results showed that Ca^{2+} are not essential for growth of *A. borkumensis* but influence its progress. Ca^{2+} probably enhance lipid accumulation at low concentration of carbon source and they are probably as well connected with the mobilisation of lipid bodies, especially in the starvation period. Contrary, our results denied the theory of calcium detoxification by its location on the surface of lipid bodies as the mutant deficient in lipid bodies formation was able to growth in the carbon-rich medium.

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YSF–81
Energetic metabolism of myelinated axons: a new trophic role for myelin sheath

S. Ravera[1], I. Panfoli[1], D. Calzia[1], M. G. Aluigi[1], P. Bianchini[2], A. Diaspro[2], G. Mancardi[3] and A. Morelli[1]
[1]*Biology Department, University of Genoa, Genova, ITALY,* [2]*Physics Department, University of Genoa, Genova, ITALY,* [3]*Neurology Opthalmology and Genetic Department, University of Genoa, Genova, ITALY*

Background: Neuronal functioning requires a lot of energy. Brain consumes more than 20% of the oxygen and glucose of the whole organism. This is quite surprising because brain mitochondrial density is lower than in other organs with less energy demand. Also, myelin sheath, the multilayered membrane allowing the nerve to transmit its impulses rapidly, exerts an as yet unexplained neuro-trophic role. In fact, in demyelinising diseases, like in Multiple Sclerosis, a lowering of conduction speed but also an axonal necrosis, is observed.
Objectives: Aiming at contributing to the understanding of the causes of the axonal degeneration consequent to myelin loss, our principal objective was to demonstrate that the electron transport chain (ETC) is functional in myelin, to carry out oxidative phosphorylation, for ATP supply to the axon.
Methods: Experiments were conducted on isolated myelin vesicles (IMV), obtained according to the method of Norton and Poduslo. Both an imaging and a biochemical approach were utilized. Transmission Electron and confocal microscopy as well as oxymetric, fluorimetric, luminometric and Semiquantitative Western Blotting (WB) analyses were performed.
Results: We observed that IMV: (i) are able to consume oxygen with NADH and Succinate as respiring substrates (ii) display a proton gradient across their surface; (iii) contain F_0–F_1 ATP Synthase and the ETC complexes, which are catalytically active. Mitochondrial contamination, as assessed by semiquantitative WB with antibodies against proteins typical of the mitochondria, was found to be negligible in IMV.
Conclusion/Application to practice: Data suggest that the whole redox chain is present in myelin sheath and that it is catalytically active in aerobic ATP production, which may be pivotal to the high energy demands of axons. This basic study will shed light on the aetiophatogenesis of many demyelinising diseases.

YSF–82
Molecular biosensor for *in vitro* cAMP measurements

R. Reinart and A. Rinken
Bioorganic Chemistry, Chemistry, Tartu, ESTONIA

Cyclic adenosine monophosphate (cAMP) is an important second messenger involved in neural signal transmission. The level of cAMP reflects the state of certain enzymes and other proteins found inside the cell or in the cell membrane, including seven trans-membrane receptors. Measuring level of cAMP can therefore give us valuable information about receptor functionality. Currently, immunological detection is the method of choice for measuring cAMP *in vitro*. During the recent years less labour-intensive and more flexible fluorescence methods have been developed. The most approved methods utilize certain cellular proteins as sensor molecules for detection of cAMP both *in vivo* and *in vitro*. Protein kinase A (PKA) and Epac are proteins specifically binding cAMP in various cell types. Nikolaev *et al.* [JBC (2004), 279, 34215] have developed a molecular biosensor, a genetically modified Epac protein containing a pair of fluorescence proteins, CFP and YFP. The monomolecular structure and ease of production of the Epac sensor protein have made it our tool of choice in cAMP measurements. Our aim is to apply the Epac-cAMP biosensor in a form of a purified protein for measuring cAMP level in various biological samples including tissue homogenates to determine dopamine D_1 and adenosine A_{2A} receptor activation. Therefore Epac-cAMP sensor protein was tagged with a 8 amino acid long Streptag-sequence, which allows highly selective purification of the protein from a crude cell lysate by using commercially available Streptactin affinity columns. For expression of the sensor protein in Sf9 cells we use baculovirus expression vector system which is cost-effective and allows fast production of the protein of interest with all the necessary folding and post-translational modifications